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Temperature and photoperiod influence prey consumption and utilization by two sympatric *Coccinella* species (Coleoptera: Coccinellidae) in conspecific and heterospecific combinations

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Abstract:【Aim】Indian subcontinent is amongst the vulnerable geographical landscapes of the world. A slight fluctuation in its climatic conditions may adversely influence its seasonal cycles and may cause a tremendous outbreak of aphids in its agroecosystems. *Coccinella septempunctata* L. and *C. transversalis* Fab. (Coleoptera: Coccinellidae) are the two sympatric aphidophagous ladybirds that abundantly inhabit the subcontinent.【Methods】The present *ex-situ* study has therefore been designed to explore how the two ladybirds would respond to increasing temperature (15, 20, 25, 30 and 35℃) and photoperiod (8L:16D, 12L:12D and 16L:8D) when allowed to feed together on a common prey resource (pea aphid) in conspecific and/or heterospecific combinations.【Results】The results revealed that 2-predator conspecific/heterospecific combinations of both the ladybirds exhibited antagonistic effects at five different temperatures and three different photoperiodic conditions. Despite of antagonistic effects, predators in conspecific/heterospecific combinations consumed, converted and utilized the prey biomass maximally at 25℃ and under long (16L:8D) photoperiodic conditions. However, their consumption rates, conversion efficiencies and growth rates were highest in heterospecific combinations. The 4th instar larvae utilized the prey biomass more efficiently to their own biomass at five different temperatures, while the adult females did the same at three different photoperiodic conditions.【Conclusion】It may therefore be inferred that increasing temperature and photoperiodic conditions might not stop the ladybirds from interacting antagonistically in conspecific and heterospecific combinations, but at 25℃ and under long (16L:8D) photoperiodic conditions the prey consumption and utilization efficiency of the interacting ladybirds would be optimum.

Key words: Ladybirds; *Coccinella septempunctata*; *Coccinella transversalis*; conversion efficiency; growth rate; predatory interactions; temperature; photoperiod

1 INTRODUCTION

The Indian subcontinent is a peninsular region in south-central Asia. It has a monsoon type climate, characterized by a distinct seasonal pattern – the cold weather season (November – February), the hot weather season (March – May), the advancing monsoon (June – September) and the retreating monsoon (October – November). The annual seasonal variation in temperature and/or photoperiod occurs in deserts, plains and mountains of the Indian subcontinent while the coastal areas do not experience much variation in climatic conditions. Thus, the Indian subcontinent may be considered amongst the vulnerable geographical landscapes of the world since a slight fluctuation in climatic conditions may adversely influence the seasonal cycles of the subcontinent.

Over the last 100 years, increase in mean temperature of the earth by about 1℃ (IPCC, 2007) has favoured tremendous outbreak of aphids as phytophagous pests (Batalden *et al.*, 2007; Parmesan, 2007; Trnka *et al.*, 2007) in the agroecosystems of Indian subcontinent. *Coccinella septempunctata* L. and *C. transversalis* Fab. (Coleoptera: Coccinellidae) are the two sympatric natural predators of aphid species that abundantly inhabit the subcontinent. *C. septempunctata* is ubiquitous with wide tolerance of different climatic conditions and able to forage in a wide range of habitats (Omkar and Pervez, 2002; Hodek and Michaud, 2008; Hodek *et al.*, 2012). It is a large ladybird, prominent in Indian agroecosystems where it is considered to be an indigenous species (Omkar and Bind, 1993; Omkar and Srivastava, 2003). *C. transversalis* is another large species of Oriental ladybird, native to India and found mainly in south

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Asia (Omkar and Bind, 1993). Since both ladybirds have an immense biocontrol potential of numerous aphid pests, it is therefore expected that when their most voracious stages (4th instar/adult females) would be released together in agricultural fields they would regulate the aphid infestation from the agricultural fields.

As mentioned above, slight increase in annual temperature amplifies the growth of aphid species in Indian subcontinent; there is therefore a need to explore whether this climatic change would increase the predation potential of ladybirds against aphid outbreaks. Earlier studies on climate change were mainly restricted to evaluate the effect of temperature and/or photoperiod on predation rates and/or growth and development and/or reproduction of individual ladybirds (Soares *et al.*, 2003; Berkvens *et al.*, 2008; Wang *et al.*, 2013, 2014; Tan *et al.*, 2014); but none of them analyzed the effect of temperature and/or photoperiod on predatory interactions (synergistic, additive or antagonistic) between the ladybirds.

The present study has therefore been designed *ex-situ* to evaluate the predatory interactions between *C. septempunctata* and *C. transversalis* in 2-predator conspecific/heterospecific larval/adult combinations at different temperatures and photoperiods. It is expected that the study would answer questions like: whether rising temperature/photoperiod would weaken predatory interactions between the ladybirds? Whether the prey consumption, conversion and utilization capacity of the ladybirds, in terms of their combined consumption rates, conversion efficiencies and growth rates, would increase with increase in temperature/photoperiod when they collectively feed on a common prey resource? Whether the release of conspecific ladybirds would control aphid infestation better than heterospecific ladybirds? And whether the adult stage of ladybirds would suppress aphids better than larval stages? It is also expected that the findings would be helpful in formulating predatory combinations that might successfully control the pea aphid infestation in agroecosystems of Indian subcontinent at broad ranges of temperatures and photoperiods.

In the present study, multiplicative risk model has been used to analyze predatory interactions. Additive models normally generate higher or unrealistic "expected prey consumption" and cannot correct for prey depletion in additive experimental designs, and in such cases multiplicative risk model is a more appropriate null model for the prey response variables that are usually measured (*e. g.*, proportion of prey consumed or surviving; see Sih *et al.*, 1998, and Mills, 2006). This model has also been used in some recent studies to evaluate predatory interactions (Omkar *et al.*, 2014; Kumar *et al.*, 2014).

2 MATERIALS AND METHODS

2.1 Stock culture

Forty adults, each of *C. septempunctata* (C7) and *C. transversalis* (Ct), were collected from the fields of Lucknow, India (26°50′N, 80°54′E). They were paired and reared for three generations in plastic Petri dishes (14.5 cm × 1.5 cm) under constant abiotic conditions (27 ± 2℃, 65% ± 5% RH, and photoperiod of 14L : 10D) in BOD incubators (Yorco Super Deluxe, YSI-440, New Delhi, India). They were provided with *ad libitum* pea aphid, *Acyrthosiphon pisum* (Harris) (Homoptera: Aphididae) reared on broad bean, *Vicia faba* L. (Fabaceae) and maintained in polyhouse (22 ± 1℃, 65% ±5% RH and 14L: 10D photoperiod). The pea aphid, *A. pisum* has been used as a common prey resource in this study since it is cosmopolitan, tolerates high temperatures (~ 40℃) and feeds on numerous host plants (*viz.* pea, beans, alfalfa, and other legumes) (Harmon *et al.*, 2009).

Wild ladybirds from the fields were frequently added to the established stock to prevent inbreeding. The eggs laid were collected every 24 h, kept in BOD incubators, observed for hatching and the requisite stages obtained were used in the experiments. Being the most voracious predatory stages, the 4th instar larvae (12 h after moulting) and 10-day old unmated adult females were used in the experiments.

Ten-day old adult females (C7 and Ct) were selected for experiments because they are: (i) still in pre-oviposition phase when fed on *A. pisum* and kept at optimum temperature and photoperiod (Bista and Omkar, 2013), (ii) more voracious than middle aged (Mishra *et al.*, 2012) and old females, and (iii) probably less susceptible to environmental stress than younger females. A preliminary trial was also conducted using 10-day old unmated females of C7 ($n = 20$) and Ct ($n = 20$) to ensure that they did not lay eggs. For this they were kept individually in Petri dishes for 24 h, provided with abundant prey and kept at 27 ± 2℃ and under a 14L: 10D photoperiod in BOD incubators. Petri dishes were checked every hour for egg deposition.

The unmated females were selected because: (i) they do not lay eggs and would avoid errors due to variations in the reproductive performance of these ladybirds (Omkar *et al.*, 2014), (ii) mated females might consume their own eggs or those of other females resulting in a reduced consumption of prey and would thus provide an unrealistic estimate of biomass of prey that can be consumed, and (iii) unmated females are more voracious than mated females (Rhamhalinghan, 1987).

2.2 Experimental design

2.2.1 Effect of temperature: Prior to the commencement of experiment, the 4th instar larvae (12 h after moulting) and 10-day-old unmated adult females were starved for 12 h and weighed (Sartorius CP225-D; 0.01 mg precision). They were then provided with 50 mg of intermediate instars of *A. pisum* in Petri dishes, and kept singly for the next 24 h under one of the following temperatures: (i) 15 ± 2℃, (ii) 20 ± 2℃, (iii) 25 ± 2℃, (iv) 30 ± 2℃, and (v) 35 ± 2℃. The used five temperatures best describe the seasonal cycles of the Indian sub-continent. Relative humidity and photoperiod remained similar to stock culture. After 24 h, the predators and remaining aphids were separated and weighed. There were 10 replicates per temperature per predatory stage per ladybird species, totaling 100 4th instar larvae and 100 adult females.

To evaluate the predatory interactions (synergistic, additive or antagonistic) between the ladybird species, 2-predator conspecific (C7 + C7, Ct + Ct) and heterospecific (C7 + Ct) 4th instar/adult female combinations were formed at each of the above five temperatures along with 100 mg (~optimum prey biomass to avoid cannibalism/intraguild predation; Kumar *et al.*, 2014) aphid mass and maintained at the above mentioned photoperiod and relative humidity for 24 h. The conspecific individuals were marked with a marker pen for visual identification before releasing them in the experimental arena. After 24 h, the remaining aphids were also separated and weighed. The 4th instar larvae/adult females were weighed individually prior to and post experiment. Each treatment was replicated ten times per temperature and per predatory stage (the operation on ten replications was done in single time duration); totaling 150 4th instar larvae and 150 adult females.

To assess the natural reduction in aphid biomass when kept at five different temperatures (if any) in absence of predators, the weighed biomass, *viz.* 50 and 100 mg of *A. pisum* was placed in Petri dishes and kept under: (i) 15 ± 2℃, (ii) 20 ± 2℃, (iii) 25 ± 2℃, (iv) 30 ± 2℃, and (v) 35 ± 2℃ for 24 h, reweighed and considered as controls. The average loss of biomass, if any, based on 5 replicates per standard aphid biomass per temperature, was used to standardize the data on consumption prior to calculating the various parameters.

2.2.2 Effect of photoperiod: While evaluating the effect of photoperiod on predatory interactions (synergistic, additive or antagonistic) between the ladybird species, either singly (totaling 60 4th instar larvae/60 adult females) or in 2-predator conspecific/heterospecific combinations (totaling 90 4th instar larvae and 90 adult females), the experimental protocol was similar to the previous experiment, except that temperature was 25 ± 2℃ as recorded optimum in the previous experiment, and photoperiodic conditions were any one of the following: (i) 8L∶16D, (ii) 12L∶12D, and (iii) 16L∶8D. There were 10 replicates per photoperiod per predatory stage for each ladybird species.

2.3 Statistical analysis

The data were checked for normal distribution using Kolmogorov-Smirnoff test for normality and Bartlett's test for homogeneity of variances prior to further analysis. The statistical analysis of the data was done to find out the: (1) predatory interactions in conspecific and heterospecific combinations at the five different temperatures/three different photoperiods, and (2) variations in conversion efficiencies and growth rates in different predatory combinations at these temperatures/photoperiods.

2.3.1 Multiplicative risk model and predatory interactions: The expected/observed prey consumption within 24 h in each predatory combination was assessed by multiplicative risk model [using equations (1), (2), (3) and (4)] to correct for prey depletion in additive experimental designs (Soluk and Collins, 1988; Sih *et al.*, 1998).

Proportion of prey consumption by predator 'a' or 'b' alone (Pa or Pb) = Prey consumption (mg) by predator 'a' or 'b' alone/Prey biomass (mg) provided to predator 'a' or 'b' alone.

Proportions of prey consumption by larval predators at different temperatures were:

(i) 15 ± 2℃: Pa (C7) = 0.47 ± 0.01; Pb (Ct) = 0.48 ± 0.01;

(ii) 20 ± 2℃: Pa (C7) = 0.56 ± 0.01; Pb (Ct) = 0.60 ± 0.02;

(iii) 25 ± 2℃: Pa (C7) = 0.73 ± 0.04; Pb (Ct) = 0.71 ± 0.04;

(iv) 30 ± 2℃: Pa (C7) = 0.55 ± 0.02; Pb (Ct) = 0.48 ± 0.01;

(v) 35 ± 2℃: Pa (C7) = 0.50 ± 0.01; Pb (Ct) = 0.41 ± 0.01.

Proportions of prey consumption by adult predators at different temperatures were:

(i) 15 ± 2℃: Pa (C7) = 0.49 ± 0.01; Pb (Ct) = 0.51 ± 0.02;

(ii) 20 ± 2℃: Pa (C7) = 0.58 ± 0.01; Pb (Ct) = 0.61 ± 0.02;

(iii) 25 ± 2℃: Pa (C7) = 0.86 ± 0.04; Pb

(Ct) = 0.88 ± 0.04;

(iv) 30 ± 2℃: *Pa* (C7) = 0.65 ± 0.02; *Pb* (Ct) = 0.53 ± 0.01;

(v) 35 ± 2℃: *Pa* (C7) = 0.53 ± 0.01; *Pb* (Ct) = 0.50 ± 0.01.

Proportions of prey consumption by larval predators at different photoperiods were:

(i) 8L∶16D: *Pa* (C7) = 0.47 ± 0.01; *Pb* (Ct) = 0.48 ± 0.02;

(ii) 12L∶12D: *Pa* (C7) = 0.65 ± 0.04; *Pb* (Ct) = 0.67 ± 0.05;

(iii) 16L∶8D: *Pa* (C7) = 0.73 ± 0.04; *Pb* (Ct) = 0.71 ± 0.04.

Proportions of prey consumption by adult predators at different photoperiods were:

(i) 8L∶16D: *Pa* (C7) = 0.78 ± 0.03; *Pb* (Ct) = 0.71 ± 0.04;

(ii) 12L∶12D: *Pa* (C7) = 0.86 ± 0.04; *Pb* (Ct) = 0.70 ± 0.02;

(iii) 16L∶8D: *Pa* (C7) = 0.89 ± 0.04; *Pb* (Ct) = 0.88 ± 0.01.

Expected proportion of prey consumption = $1 - [(1 - Pa)(1 - Pb)]$

where *Pa* and *Pb* are the proportion of prey consumption by predators ‘a’ and ‘b’ alone, respectively.

Observed proportion of prey consumption (in 2-predator conspecific/heterospecific combinations) = Combined prey consumption (mg) by two predator species/Prey biomass (mg) provided;

Expected/Observed prey consumption (%) = Expected/Observed proportion of prey consumption × 100.

Variation in expected and observed prey consumption in each predatory combination was analyzed using one-way ANOVA. All percent data were subjected to arcsine square root transformation prior to ANOVA. For assessing the influence of temperature/photoperiod, predatory stage and predatory combinations on prey consumption, the data were subjected to three-way ANOVA followed by Tukey's *post hoc* comparison of means.

2.3.2 Conversion efficiencies and growth rates: The conversion efficiency and growth rate in each predatory combination (4th instar larvae/adult females) were calculated using the following formulae:

Conversion efficiency (modified after Dixon, 2000) = Change in combined biomass (mg) of both predators in predatory combinations/Biomass (mg) of prey consumed by both predators in predatory combinations;

Growth rate (per day) (modified after Waldbauer, 1968) = Change in combined biomass (mg) of both predators in predatory combinations/(Duration of feeding period × Combined mean biomass (mg) of both predators).

Variations in conversion efficiencies and growth rates were analyzed using three-way ANOVA with temperature/photoperiod, predatory stage (the 4th instar larva and female adult) and predatory combinations (C7 + C7, Ct + Ct and C7 + Ct) as independent factors and conversion efficiencies and growth rates as dependent factors, followed by Tukey's *post hoc* comparison of means. All statistical analyses were performed using MINITAB 16 (Minitab Inc., State College, Pennsylvania, USA).

3 RESULTS

3.1 Effect of temperature on prey consumption and utilization

3.1.1 Larval combinations: Multiplicative risk model analysis revealed that the 2-predator conspecific and heterospecific combinations showed antagonistic effect at all temperatures tested. Thus, in the larval conspecific/heterospecific combinations, observed prey consumption by the predators was lower than the expected prey consumption when kept at different temperatures (Table 1).

3.1.2 Adult combinations: The adult predators in conspecific (C7 + C7 and Ct + Ct) and heterospecific (C7 + Ct) combinations showed antagonistic effects at all temperatures (Table 2).

Results of three-way ANOVA further revealed that the observed prey consumption (= prey consumption percentage) by predators in combinations was significantly influenced by the independent factors, *viz.* temperature, predatory combinations ($F = 48.68$, $P < 0.0001$, $df = 2, 299$) and the stage of the predators ($F = 362.73$, $P < 0.0001$, $df = 1, 299$). Also the interactions between the independent factors significantly ($F_{\text{Temperature × Predatory combinations}} = 3.10$, $P = 0.002$, $df = 8, 299$; $F_{\text{Temperature × Stage}} = 5.28$, $P < 0.0001$, $df = 4, 299$ and $F_{\text{Predatory combinations × Stage}} = 10.21$, $P < 0.0001$, $df = 2, 299$) affected the observed prey consumption of predators.

The results therefore exemplified that despite of the presence of antagonistic effects, predators had the highest prey consumption at 25℃ in conspecific/heterospecific combinations. Also at all temperatures, (i) larval/adult predators in heterospecific combinations had higher prey consumption than predators in conspecific combinations, and (ii) adult predators consumed more prey than the larval predators in their respective conspecific/heterospecific combinations.

Table 1 Effect of temperature on observed and expected prey consumption (%) showing overall predatory interactions using Multiplicative Risk Model in larval combinations of *Coccinella septempunctata* and *C. transversalis*

Temperature	Predatory Combinations	Combined prey consumption (mg)	Sum of individual prey consumptions (mg)	Multiplicative Risk Model (4th instar)			
				Observed prey consumption (%)	Expected prey consumption (%)	*F*-value (*P*-value)	Predatory interaction
15℃	C7 + C7	50.75 ± 1.44	46.80 ± 0.78	50.75 ± 1.44 Ba	71.64 ± 0.84	156.99 (<0.0001)	Antagonism
	Ct + Ct	32.42 ± 1.79	48.16 ± 1.92	32.42 ± 1.79 Aa	72.79 ± 1.92	237.57 (<0.0001)	Antagonism
	C7 + Ct	51.98 ± 3.24	43.37 ± 1.29	51.98 ± 3.24 Ba	67.78 ± 1.47	19.70 (<0.0001)	Antagonism
20℃	C7 + C7	55.38 ± 2.94	55.97 ± 1.32	55.38 ± 2.94 Bc	80.45 ± 1.16	62.79 (<0.0001)	Antagonism
	Ct + Ct	50.75 ± 1.44	59.79 ± 2.49	50.75 ± 1.44 Ab	83.27 ± 2.14	158.93 (<0.0001)	Antagonism
	C7 + Ct	68.40 ± 3.76	65.14 ± 3.26	68.40 ± 3.76 Cb	86.89 ± 2.53	16.60 (0.001)	Antagonism
25℃	C7 + C7	61.42 ± 3.11	72.74 ± 4.01	61.42 ± 3.11 Ad	91.12 ± 1.60	72.14 (<0.0001)	Antagonism
	Ct + Ct	57.84 ± 4.37	71.08 ± 4.32	57.84 ± 4.37 Ad	89.96 ± 2.99	36.79 (<0.0001)	Antagonism
	C7 + Ct	74.93 ± 2.15	72.09 ± 2.61	74.93 ± 2.15 Bc	91.60 ± 1.77	35.77 (<0.0001)	Antagonism
30℃	C7 + C7	57.32 ± 1.82	54.97 ± 1.59	57.32 ± 1.82 Ad	79.50 ± 1.37	94.66 (<0.0001)	Antagonism
	Ct + Ct	53.71 ± 3.47	48.10 ± 1.34	53.71 ± 3.47 Ac	72.91 ± 1.38	26.37 (<0.0001)	Antagonism
	C7 + Ct	72.80 ± 1.50	58.01 ± 0.93	72.80 ± 1.50 Bc	82.30 ± 0.79	31.16 (<0.0001)	Antagonism
35°C	C7 + C7	54.26 ± 1.41	49.57 ± 0.86	54.26 ± 1.41 Bb	74.50 ± 0.87	149.06 (<0.0001)	Antagonism
	Ct + Ct	50.53 ± 2.58	40.82 ± 0.87	50.53 ± 2.58 Ab	64.91 ± 1.04	26.71 (<0.0001)	Antagonism
	C7 + Ct	69.71 ± 1.87	52.49 ± 1.21	69.71 ± 1.87 Cb	77.30 ± 1.04	12.04 (0.003)	Antagonism

Values are mean ± *SE*; *F*-values significant at $P < 0.05$; *df* = 1, 19. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively. Large letters compare observed prey consumption among predatory combinations (in the same temperature) and small letters compare observed prey consumption among temperatures (among the same combination), based on Tukey's *post hoc* comparison of means.

Table 2 Effect of temperature on observed and expected prey consumption (%) showing overall predatory interactions using Multiplicative Risk Model in adult female combinations of *Coccinella septempunctata* and *C. transversalis*

Temperature	Predatory combinations	Combined prey consumption (mg)	Sum of individual prey consumptions (mg)	Multiplicative Risk Model (adult females)			
				Observed prey consumption (%)	Expected prey consumption (%)	*F*-value (*P*-value)	Predatory interaction
15℃	C7 + C7	67.60 ± 1.55	48.71 ± 0.70	67.60 ± 1.55 Aa	73.65 ± 0.72	12.53 (0.002)	Antagonism
	Ct + Ct	65.22 ± 2.34	50.54 ± 2.10	65.22 ± 2.34 Aa	75.14 ± 2.11	9.92 (0.006)	Antagonism
	C7 + Ct	71.71 ± 1.32	52.07 ± 1.44	71.71 ± 1.32 Ba	76.84 ± 1.43	6.94 (0.017)	Antagonism
20℃	C7 + C7	74.85 ± 0.59	58.17 ± 1.16	74.85 ± 0.59 Bc	82.38 ± 0.97	43.92 (<0.0001)	Antagonism
	Ct + Ct	72.12 ± 3.29	61.31 ± 2.22	72.12 ± 3.29 Bb	84.59 ± 1.84	10.92 (0.004)	Antagonism
	C7 + Ct	68.40 ± 3.76	68.62 ± 2.45	68.40 ± 3.76 Aa	89.61 ± 1.36	28.10 (<0.0001)	Antagonism
25℃	C7 + C7	89.92 ± 1.53	85.78 ± 4.13	89.92 ± 1.53 ABe	96.44 ± 1.77	7.76 (0.012)	Antagonism
	Ct + Ct	85.85 ± 3.18	88.00 ± 1.12	85.85 ± 3.18 Ac	98.45 ± 0.25	15.62 (0.001)	Antagonism
	C7 + Ct	91.14 ± 1.26	85.21 ± 1.32	91.14 ± 1.26 Bc	97.66 ± 0.41	24.12 (<0.0001)	Antagonism
30℃	C7 + C7	77.00 ± 0.67	65.48 ± 1.58	77.00 ± 0.67 Bd	87.86 ± 1.09	72.67 (<0.0001)	Antagonism
	Ct + Ct	71.45 ± 1.04	53.33 ± 0.47	71.45 ± 1.04 Ab	78.20 ± 0.44	35.75 (<0.0001)	Antagonism
	C7 + Ct	89.09 ± 1.26	59.57 ± 1.57	89.09 ± 1.26 Cc	83.45 ± 1.26	18.38 (<0.0001)	Antagonism
35℃	C7 + C7	71.30 ± 1.52	53.01 ± 1.16	71.30 ± 1.52 Bb	77.80 ± 1.09	12.04 (0.003)	Antagonism
	Ct + Ct	67.45 ± 1.12	50.34 ± 1.22	67.45 ± 1.12 Aa	75.20 ± 1.25	21.39 (<0.0001)	Antagonism
	C7 + Ct	74.80 ± 0.63	55.93 ± 0.97	74.80 ± 0.63 Cb	79.60 ± 0.83	21.16 (<0.0001)	Antagonism

Values are mean ± *SE*; *F*-values significant at $P < 0.05$; *df* = 1, 19. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively. Large letters compare observed prey consumption among predatory combinations (in the same temperature) and small letters compare observed prey consumption among temperatures (among the same combination), based on Tukey's *post hoc* comparison of means.

3.1.3 Conversion efficiencies and growth rates: Three-way ANOVA revealed significant influence of the independent factors, *viz.* temperature ($F = 5.51$, $P < 0.0001$, $df = 4, 299$), predatory combinations ($F = 116.97$, $P < 0.0001$, $df = 2, 299$) and predatory stage ($F = 371.54$, $P < 0.0001$, $df = 1, 299$) on the conversion efficiency. The conversion efficiency of predators within combinations differed significantly with the interactions between temperature and predatory combinations ($F = 3.66$, $P < 0.0001$, $df = 8, 299$) and between predatory combinations and predatory stage ($F = 17.55$, $P < 0.0001$, $df = 2, 299$), but not with the interaction between temperature and predatory stage ($F = 0.70$, $P = 0.594$, $df = 4, 299$).

Three-way ANOVA revealed significant influence of temperature ($F = 23.99$, $P < 0.0001$,

$df=4$, 299), predatory combinations ($F=545.91$, $P<0.0001$, $df=2$, 299) and predatory stage ($F=511.68$, $P<0.0001$, $df=1$, 299) on the growth rate. The growth rate of predators within combinations also differed significantly with the interactions between temperature and predatory combinations ($F=6.44$, $P<0.0001$, $df=8$, 299), between predatory combinations and predatory stage ($F=107.91$, $P<0.0001$, $df=2$, 299) and between temperature and predatory stage ($F=8.12$, $P<0.0001$, $df=4$, 299).

Results therefore revealed that: (i) the conversion efficiencies and growth rates of predators were highest at 25℃, and (ii) both the parameters were highest in heterospecific larval combinations (Fig. 1).

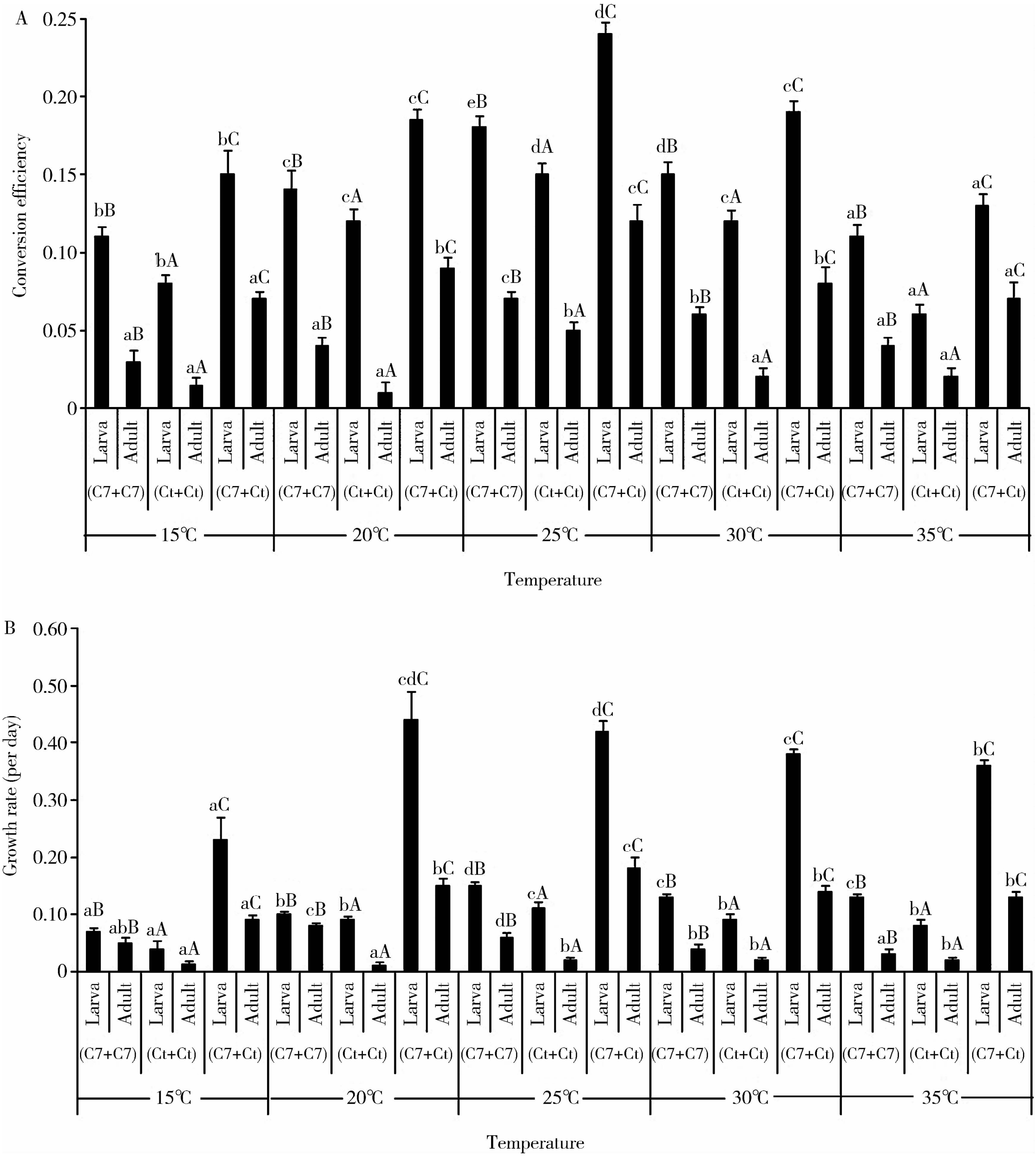

Fig. 1 Effect of temperature on conversion efficiency (A) and growth rate (B) of larval/adult predators within conspecific and heterospecific combinations of *Coccinella septempunctata* and *C. transversalis*

Values are mean ± *SE*; small and large letters represent comparison of means between temperature and predatory combinations, respectively. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively.

3.2 Effect of photoperiod on prey consumption and utilization

3.2.1 Larval combinations: The predator larvae showed antagonistic effects within conspecific (C7 + C7 and Ct + Ct) and heterospecific (C7 + Ct) combinations at three photoperiods tested so far. Thus, the observed prey consumption was lower than the expected prey consumption when the predator larvae were exposed to different photoperiodic regimes (Table 3).

Table 3 Effect of photoperiod on observed and expected prey consumption (%) showing overall predatory interactions using Multiplicative Risk Model in larval combinations of *Coccinella septempunctata* and *C. transversalis*

Photoperiod	Predatory combinations	Combined prey consumption (mg)	Sum of individual prey consumptions (mg)	Multiplicative Risk Model (4th instar)			
				Observed prey consumption (%)	Expected prey consumption (%)	*F*-value (*P*-value)	Predatory interaction
8L∶16D	C7 + C7	50.75 ± 1.44	46.80 ± 0.78	50.75 ± 1.44 Ba	71.64 ± 0.84	156.99 (<0.0001)	Antagonism
	Ct + Ct	32.42 ± 1.79	48.16 ± 1.92	32.42 ± 1.79 Aa	72.79 ± 1.92	237.57 (<0.0001)	Antagonism
	C7 + Ct	51.98 ± 3.24	43.37 ± 1.29	51.98 ± 3.24 Ba	67.78 ± 1.47	19.70 (<0.0001)	Antagonism
12L∶12D	C7 + C7	55.38 ± 2.94	65.00 ± 3.53	55.38 ± 2.94 Bb	86.63 ± 2.29	70.35 (<0.0001)	Antagonism
	Ct + Ct	50.75 ± 1.44	67.20 ± 4.59	50.75 ± 1.44 Ab	87.34 ± 3.17	110.56 (<0.0001)	Antagonism
	C7 + Ct	68.40 ± 3.76	65.14 ± 3.26	68.40 ± 3.76 Cb	86.89 ± 2.53	16.60 (0.001)	Antagonism
16L∶8D	C7 + C7	61.42 ± 3.11	72.74 ± 4.01	61.42 ± 3.11 Ac	91.12 ± 1.60	72.14 (<0.0001)	Antagonism
	Ct + Ct	57.84 ± 4.37	71.08 ± 4.32	57.84 ± 4.37 Ac	89.96 ± 2.99	36.79 (<0.0001)	Antagonism
	C7 + Ct	74.93 ± 2.15	72.09 ± 2.61	74.93 ± 2.15 Bc	91.60 ± 1.77	35.77 (<0.0001)	Antagonism

Values are mean ± *SE*; *F*-values significant at $P < 0.05$; $df = 1, 19$. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively. Large letters compare observed prey consumption among predatory combinations (in the same photoperiod) and small letters compare observed prey consumption among photoperiods (among the same combination), based on Tukey's *post hoc* comparison of means.

3.2.2 Adult combinations: The adult predators within 2-predator conspecific and heterospecific combinations showed antagonistic effects at three photoperiods (Table 4). Three-way ANOVA revealed that the observed prey consumption (= prey consumption percentage) was significantly influenced by the photoperiodic regimes ($F = 38.17$, $P < 0.0001$, $df = 2, 179$), predatory combinations ($F = 12.98$, $P < 0.0001$, $df = 2, 179$), predatory stage ($F = 195.47$, $P < 0.0001$, $df = 1, 179$) and their interaction ($F = 2.56$, $P = 0.041$, $df = 4, 179$).

Results therefore revealed that despite optimum temperature and antagonistic effects, (i) prey consumption by predators was highest at 16L∶8D, (ii) predators of heterospecific combinations consumed higher prey biomass than the conspecific combinations, and (iii) adult predators consumed higher prey biomass than the larval predators within their respective conspecific/heterospecific combinations.

3.2.3 Conversion efficiencies and growth rates: Three-way ANOVA revealed that the conversion efficiency of predators within combinations differed significantly with photoperiod ($F = 95.41$, $P < 0.0001$, $df = 2, 179$), predatory combinations ($F = 57.34$, $P < 0.0001$, $df = 2, 179$) and the predatory stage ($F = 40.55$, $P < 0.0001$, $df = 1, 179$). However, the interaction between the three independent factors, *viz.* photoperiod, predatory combinations and predatory stage was not significant for the conversion efficiency ($F = 1.91$, $P = 0.111$, $df = 4, 179$).

Three-way ANOVA further revealed that the growth rate of predators within combinations differed significantly with photoperiod ($F = 93.37$, $P < 0.0001$, $df = 2, 179$), predatory combinations ($F = 53.28$, $P < 0.0001$, $df = 2, 179$) and the predatory stage ($F = 75.30$, $P < 0.0001$, $df = 1, 179$). Also the interaction between the three independent factors was significant for the growth rate ($F = 2.50$, $P = 0.045$, $df = 4, 179$).

Results therefore exemplified that despite optimum temperature, (i) conversion efficiencies and growth rates were highest at 16L∶8D, and (ii) both parameters were highest within heterospecific adult combinations (Fig. 2).

Table 4 Effect of photoperiod on observed and expected prey consumption (%) showing overall predatory interactions using Multiplicative Risk Model in adult female combinations of *Coccinella septempunctata* and *C. transversalis*

Photoperiod	Predatory combinations	Combined prey consumption (mg)	Sum of individual prey consumptions (mg)	Multiplicative Risk Model (adult females)			
				Observed prey consumption (%)	Expected prey consumption (%)	*F*-value (*P*-value)	Predatory interaction
8L∶16D	C7 + C7	73.83 ± 7.59	70.40 ± 4.22	73.83 ± 7.59 Aa	98.96 ± 0.43	10.92 (0.004)	Antagonism
	Ct + Ct	71.50 ± 5.92	68.62 ± 2.45	71.50 ± 5.92 Aa	94.39 ± 1.50	14.03 (0.001)	Antagonism
	C7 + Ct	74.93 ± 2.15	71.08 ± 4.32	74.93 ± 2.15 Aa	89.96 ± 2.99	16.62 (0.001)	Antagonism
12L∶12D	C7 + C7	84.30 ± 1.00	85.21 ± 1.32	84.30 ± 1.00 Bb	97.44 ± 2.07	32.64 (<0.0001)	Antagonism
	Ct + Ct	72.12 ± 3.29	78.24 ± 3.12	72.12 ± 3.29 Aa	89.63 ± 2.70	16.88 (0.001)	Antagonism
	C7 + Ct	68.40 ± 3.76	85.78 ± 4.13	69.40 ± 3.76 Aa	89.61 ± 1.36	28.10 (<0.0001)	Antagonism
16L∶8D	C7 + C7	89.92 ± 1.53	89.40 ± 4.00	89.92 ± 1.53 Bc	96.44 ± 1.77	7.76 (0.012)	Antagonism
	Ct + Ct	85.85 ± 3.18	88.00 ± 1.12	85.85 ± 3.18 Ab	98.45 ± 0.25	15.62 (0.001)	Antagonism
	C7 + Ct	91.14 ± 1.26	92.18 ± 2.18	91.14 ± 1.26 Bb	97.66 ± 0.41	24.12 (<0.0001)	Antagonism

Values are mean ± *SE*; *F*-values significant at $P < 0.05$; $df = 1, 19$. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively. Large letters compare observed prey consumption among predatory combinations (in the same photoperiod) and small letters compare observed prey consumption among photoperiods (among the same combination), based on Tukey's *post hoc* comparison of means.

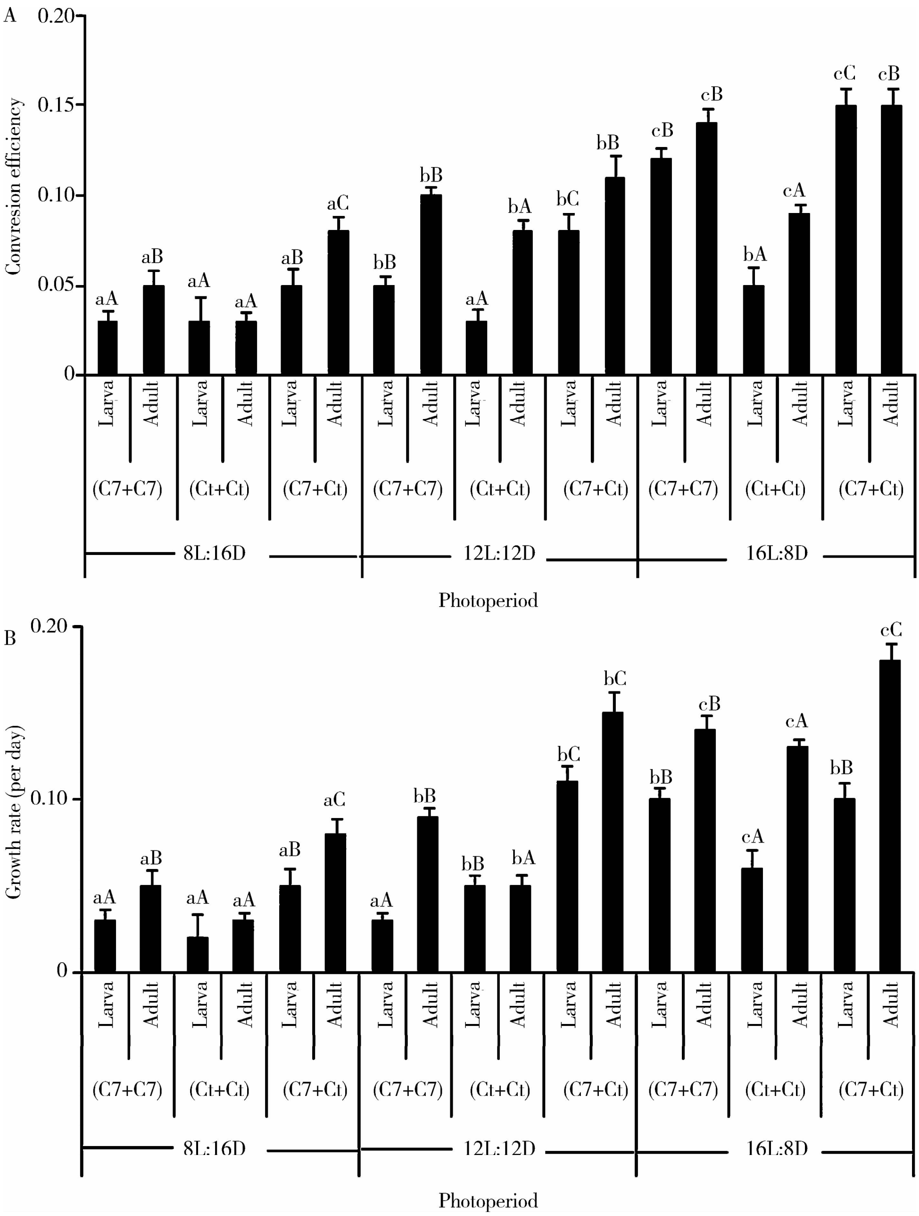

Fig. 2 Effect of photoperiod on the conversion efficiency (A) and growth rate (B) of larval/adult predators within conspecific and heterospecific combinations of *Coccinella septempunctata* and *C. transversalis*

Values are mean ± *SE*; small and large letters represent comparison of means between photoperiod and predatory combinations, respectively. C7 and Ct represent *C. septempunctata* and *C. transversalis*, respectively.

4 DISCUSSION

In the present study, predators within conspecific (*C. septempunctata* + *C. septempunctata* and *C. transversalis* + *C. transversalis*) and heterospecific (*C. septempunctata* + *C. transversalis*) combinations showed antagonistic effects at five different temperatures and three different photoperiodic conditions. This may be due to the predator-predator interactions within the combinations. In the presence of other predator(s), individual predator probably consumes lower prey biomass than its individual predation rate, which results in an overall lower prey consumption within different predatory combinations. The present findings are in conformity with earlier reports in ladybirds (Müllller and Godfray, 1999a, 1999b; Snyder and Ives, 2001; Kaplan and Eubanks, 2002; Denno and Finke, 2006; Mills, 2006; Nóia *et al.*, 2008; Hodek *et al.*, 2012; Omkar *et al.*, 2014; Kumar *et al.*, 2014). Despite of antagonistic effects, predators within combinations have shown highest consumption rates, conversion efficiencies and growth rates at 25℃ and under long (16L∶8D) photoperiodic conditions. It therefore appears that 25℃ and long day lengths are the best suitable abiotic conditions that possibly promote the ladybirds in combination to consume, convert and utilize the prey biomass maximally to their own biomass. Earlier, the 4th instar larvae and adult females of ladybird, *Cheilomenes sexmaculata* (F.) increased their prey consumption rates with increasing temperature and photoperiod (Wang *et al.*, 2013). Similarly, in ladybird, *Hippodamia variegata* (Goeze), prey consumption rate increased with prolonged day length (Tan *et al.*, 2014).

Further, heterospecific combinations have demonstrated higher prey consumption, conversion and utilization than the conspecific combinations at different temperatures/photoperiods possibly because of the relatively low predator-predator interference. The conspecific predators are of similar size, occupy the same ecological niche, and have similar defence mechanism, vigour and energy/food requirements. As a result they severely compete for the food and space. On the other hand, the heterospecific predators have relatively dissimilar food requirements, defence mechanisms, feeding habits and body size, which result in poor competition owing to low niche overlap than the conspecific predators. Consequently the former spend more time in prey consumption than the latter. In contrast, the predators in conspecific combinations appear to aggressively compete for the food and space that enhances their metabolic costs. But their prey consumption does not seem to be sufficient to meet their higher metabolic costs that probably results in their lower conversion efficiencies and growth rates. The findings on lower predatory interference within the heterospecific combinations support earlier reports in ladybirds (Connell, 1983; Evans, 1991; Omkar and Pervez, 2011; Kumar *et al.*, 2014).

In the present study, higher prey consumption by the adult predators at different temperatures and photoperiods than the larval predators reveals that the predatory interactions are probably weak in adult ladybirds. They may therefore spend most of their time on predation rather than interacting with each other and may show higher prey consumption than larvae to fulfill their: (i) higher metabolic costs associated with flight, searching of mates and oviposition sites, and (ii) high energy requirements needed for reproduction and oviposition.

Despite of lower prey consumption the larval conspecific/heterospecific combinations have shown higher conversion efficiencies and growth rates than adult combinations at different temperatures. Since larvae have higher conversion efficiencies and growth rates than the adults (Mishra *et al.*, 2011), it appears that larval tendency to efficiently exploit and utilize prey biomass to their own biomass remains unaffected at different temperatures. And when kept in combinations at different temperatures, larvae possibly compensate for their increased nutritional needs by exploiting the prey biomass more efficiently than the adults. In contrast, adults have exhibited higher consumption rates, conversion efficiencies and growth rates than the larvae under different photoperiodic conditions, probably owing to strong predatory interactions within larval combinations that subjugate their higher prey utilization efficiency.

The present *ex-situ* study thus reveals that increasing temperature and photoperiodic conditions might not stop the ladybirds from interacting antagonistically within conspecific and heterospecific combinations; but at 25℃ and under long (16L: 8D) photoperiodic conditions prey consumption and utilization efficiency of the interacting ladybirds would be optimum. The study also exemplifies that increase in mean annual temperature of the Indian subcontinent from 15 to 25℃ (in winters and monsoons) would possibly allow conspecific/heterospecific ladybirds to efficiently consume, convert and utilize prey biomass to their own biomass when released collectively. But, the short day lengths of winters/monsoons might reduce their prey consumption and utilization ability to certain extent. Further rise in temperature above 25℃ (in summers) would hopefully enable conspecific/heterospecific ladybirds to consume and utilize less prey biomass; but long day lengths in summers might provide them an opportunity to improve their prey consumption and utilization ability. However, field-based studies are still needed to draw broad based and more meaningful conclusions.

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References

Batalden RV, Oberhauser K, Peterson AT, 2007. Ecological niches in sequential generations of eastern North American Monarch butterflies (Lepidoptera: Danaidae): the ecology of migration and likely climate change implications. *Environ. Entomol.*, 36: 1365 – 1373.

Berkvens N, Bonte J, Berkvens D, Tirry L, De Clercq P, 2008. Influence of diet and photoperiod on development and reproduction of European populations of *Harmonia axyridis* (Pallas) (Coleoptera: Coccinellidae). *BioControl*, 53: 211 – 221.

Bista M, Omkar, 2013. Effects of body size and prey quality on the reproductive attributes of two aphidophagous Coccinellidae (Coleoptera) species. *Can. Entomol.*, 145(5): 566 – 576.

Connell JH, 1983. On the prevalence and relative importance of interspecific competition: evidence from field experiments. *Am. Nat.*, 122: 661 - 696.

Denno RF, Finke DL, 2006. Multiple predator interactions and food-web connectance - implications for biological control. In: Brodeur J, Boivin G eds. Trophic and Guild Interactions in Biological Control. Springer, Dordrecht, The Netherlands. 45 - 70.

Dixon AFG, 2000. Insect Predator-Prey Dynamics, Ladybird Beetles and Biological Control. Cambridge University Press, Cambridge.

Evans EW, 1991. Intra versus interspecific interactions of ladybeetles (Coleoptera: Coccinellidae) attacking aphids. *Oecologia*, 87: 401 - 408.

Harmon JP, Moran NA, Ives AR, 2009. Species response to environmental change: impacts of food web interactions and evolution. *Science*, 323(5919): 1347 - 1350.

Hodek I, Michaud JP, 2008. Why is *Coccinella septempunctata* so successful? *Eur. J. Entomol.*, 105: 1 - 12.

Hodek I, Van Emden HF, Honek A, 2012. Ecology and Behaviour of the Ladybird Beetles (Coccinellidae). Blackwell Publishing Limited, U. K.

IPCC (Intergovernmental Panel on Climate Change), 2007. Climate change 2007: the scientific basis. In : Solomon S, Qin D, Manning M, Chen Z, Marquis M, Averyt KB, Tignor M, Miller HL eds. Contributions of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change: "The Physical Science Basis". Cambridge University Press, Cambridge. 1 - 18.

Kaplan I, Eubanks MD, 2002. Disruption of cotton aphid (Homoptera: Aphididae)-natural enemy dynamic by red imported fire ants (Hymenoptera: Formicidae). *Environ. Entomol.*, 31: 1175 - 1183.

Kumar B, Mishra G, Omkar, 2014. Functional response and predatory interactions in conspecific and heterospecific combinations of two congeneric species (Coleoptera: Coccinellidae). *Eur. J. Entomol.*, 111(2): 257 - 265.

Mills N, 2006. Interspecific competition among natural enemies and single versus multiple introductions in biological control: progress in biological control. In: Brodeur J, Boivin G eds. Trophic and Guild Interactions in Biological Control. Springer, The Netherlands. 191 - 220.

Mishra G, Kumar B, Shahid M, Singh D, Omkar, 2011. Evaluation of four co-occurring ladybirds for use as biocontrol agents of the pea aphid, *Acyrthosiphon pisum* (Homoptera: Aphididae). *Biocontr. Sci. Techn.*, 21: 991 - 997.

Mishra G, Omkar, Kumar B, Pandey G, 2012. Stage and age-specific predation in four aphidophagous ladybird beetles. *Biocontr. Sci. Techn.*, 22: 463 - 476.

Müller HCB, Godfray HCJ, 1999a. Indirect interactions in aphid-parasitoid communities. *Res. Popul. Ecol.*, 41: 93 - 106.

Müller HCB, Godfray HCJ, 1999b. Predators and mutualists influence the exclusion of aphid species from natural communities. *Oecologia*, 119: 120 - 125.

Nóia M, Borges L, Soares AO, 2008. Intraguild predation between the aphidophagous ladybird beetles *Harmonia axyridis* and *Coccinella undecimpunctata* (Coleoptera: Coccinellidae): the role of intra and extraguild prey densities. *Biol. Control*, 46: 140 - 146.

Omkar, Bind RB, 1993. Records of aphid-natural enemies complex of Uttar Pradesh. II. The coccinellids. *J. Adv. Zool.*, 14(2): 96 - 99.

Omkar, Mishra G, Kumar B, Singh N, Pandey G, 2014. Risks associated with tandem release of large and small ladybirds (Coleoptera: Coccinellidae) in heterospecific aphidophagous guilds. *Can. Entomol.*, 146: 52 - 66.

Omkar, Pervez A, 2002. Ecology of aphidophagous ladybird beetle, *Coccinella septempunctata* Linn. (Coleoptera: Coccinellidae): a review. *J. Aphidol.*, 16(1 - 2): 175 - 201.

Omkar, Pervez A, 2011. Functional response of two aphidophagous ladybirds searching in tandem. *Biocontr. Sci. Techn.*, 21: 101 - 111.

Omkar, Srivastava S, 2003. Comparative prey consumption and searching efficiency of ladybeetles, *Coccinella septempunctata* Linnaeus and *Coccinella transversalis* Fabricius for different aphid species. *J. Biol. Control*, 17(1): 35 - 41.

Parmesan C, 2007. Influences of species, latitudes and methodologies on estimates of phenological response to global warming. *Glob. Change Biol.*, 13: 1860 - 1872.

Rhamhalinghan M, 1987. Feeding behaviour of *Coccinella septempunctata* L. var. *confusa* Wiedemann (Coleoptera: Coccinellidae) in relation to temperature . I. Pre-oviposition period. *J. Entomol. Res.*, 11: 178 - 183.

Sih A, Englund G, Wooster D, 1998. Emergent impacts of multiple predators on prey. *Trends Ecol. Evol.*, 13(9): 350 - 355.

Snyder WE, Ives AR, 2001. Generalist predators disrupt biological control by a specialist parasitoid. *Ecology*, 82: 705 - 716.

Soares AO, Coderre D, Schanderl H, 2003. Effect of temperature and intraspecific allometry on predation by two phenotypes of *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae). *Environ. Entomol.*, 3(5): 939 - 944.

Soluk DA, Collins NC, 1988. Synergistic interactions between fish and stoneflies: facilitation and interference among stream predators. *Oikos*, 52: 94 - 100.

Tan X, Zhao J, Zhang F, Wang S, 2014. Light dependency of life traits, reproduction, locomotion, and predation in the polyphagous ladybird *Hippodamia variegata*. *Entomol. Exp. Appl.*, 152(3): 200 - 208.

Trnka M, Muškab F, Semerádová D, Dubrovskyc M, Kocmánkováa E, Žaluda Z, 2007. European corn borer life stage model: regional estimates of pest development and spatial distribution under present and future climate. *Ecol. Modell.*, 207: 61 - 84.

Waldbauer GP, 1968. The consumption and utilization of food by insect. *Adv. Insect Physiol.*, 5: 229 - 288.

Wang S, Tan XL, Guo XJ, Zhang F, 2013. Effect of temperature and photoperiod on the development, reproduction, and predation of the predatory ladybird *Cheilomenes sexmaculata* (Coleoptera: Coccinellidae). *J. Econ. Entomol.*, 106(6): 2621 - 2629.

Wang S, Wang K, Michaud JP, Zhang F, Tan XL, 2014. Reproductive performance of *Propylea japonica* (Coleoptera: Coccinellidae) under various light intensities, wavelengths and photoperiods. *Eur. J. Entomol.*, 111(3): 341 - 347.

温度和光周期影响两种同域发生瓢虫同种和异种组合时的猎物消耗量和利用率

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摘要:【目的】印度次大陆是世界上最脆弱的地理景观。气候条件的略微变动可能对其季节周期可能产生不良影响,并引起农业生态系统中蚜虫的大暴发。七星瓢虫 *Coccinella septempunctata* L. 和狭臀瓢虫 *C. transversalis* Fab. 是该次大陆上广泛分布、同域发生的两种食蚜性瓢虫。【方法】设计异地试验,来探究同种和/或异种组合时的这两种瓢虫用共同的猎物资源(豌豆蚜)饲养时,对增加的温度(15, 20, 25, 30 和 35℃)和光周期(8L: 16D, 12L: 12D 和16L: 8D)的响应。【结果】结果表明,在这 5 个不同温度和 3 个不同光周期条件下,同种或异种组合时这两种瓢虫表现出了拮抗作用。尽管表现出拮抗作用,但是同种或异种组合的两捕食动物在 25℃和长光周期(16L: 8D)条件下消耗、转化和利用的猎物生物量最大。然而,它们的猎物消耗率、转化效率和生长速率在异种组合中最高。在 5 个不同温度下,4 龄幼虫均更有效地利用猎物生物量,将其转变成自身生物量,而雌成虫在 3 个不同光周期条件下也是如此。【结论】因此可以推断,增加的温度和光周期条件可能不会阻止同种和异种组合中的瓢虫发生拮抗作用,但是在 25℃和长光周期(16L: 8D)条件下,相互作用的瓢虫的猎物消耗量和利用率为最佳。

关键词: 瓢虫; 七星瓢虫; 狭臀瓢虫; 转化效率; 生长速率; 捕食关系; 温度; 光周期

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